# Frequency and Type of Epidermal Growth Factor Receptor Mutations in African Americans with Non-small Cell Lung Cancer

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**Background:** Epidermal growth factor receptor (EGFR) mutations in non-small cell lung cancer (NSCLC) predict response to tyrosine kinase inhibitors. Mutations occur more commonly in never smokers and East Asians, but there are conflicting reports on the frequency of *EGFR* mutations in tumors from African Americans.

**Methods:** Tumors from 67 African American and 77 white participants in previous case-control studies of lung cancer were selected to determine *EGFR* mutational status. Mutation analysis was performed using the Sequenom mass array analyzer (Sequenom, San Diego, CA).

**Results:** Overall, 13.9% of the study population carried an *EGFR* mutation. *EGFR* mutations occurred in 11.9% of tumors from African Americans compared with 15.6% in whites ($p = 0.53$). All mutations found in African Americans were deletions in exon 19. The majority of mutations were found in nonsmokers among both African Americans (7/8) and whites (8/12).

**Conclusion:** These results indicate that African Americans with NSCLC harbor somatic *EGFR* mutations at a frequency similar to whites with NSCLC. Thus, clinicians should not use race as a clinical decision parameter for the use of EGFR-tyrosine kinase inhibitors.

**Key Words:** *EGFR* mutation, Race, African American, Lung cancer.

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Epidermal growth factor receptor (EGFR) is a tyrosine kinase involved in cell signaling, and somatic mutations in patients with non-small cell lung cancer (NSCLC) are predictive of response to EGFR tyrosine kinase inhibitors (TKIs), with 70 to 80% of patients deriving substantial benefit from this therapy.[1,2] Several studies have documented that most patients responding to EGFR-TKIs have mutations in the ATP-binding regions of the gene.[3–6] More than 90% of these mutations involve either a deletion in exon 19 or a point mutation (L858R) in exon 21.[7]

*EGFR* mutations occur predominantly in never smokers and are more common in patients with adenocarcinoma, East Asian ethnicity, and in women.[3–6] Less is known about the mutation frequency in other ethnic groups, and studies in African American patients have yielded conflicting results. Two studies reported that the *EGFR* mutation frequency in African Americans was about 2% compared with the reported frequency of 15 to 20% in whites.[7,8] In contrast, Riely et al.[9] reported 42.9% of the 14 tumors, which they studied from African Americans-harbored *EGFR* mutations.

The decision to offer EGFR testing is usually determined by whether the patient matches the clinical profile of an individual who would be likely to harbor a mutation. The purpose of this investigation was to examine the frequency and characteristics of *EGFR* mutations in a cohort of African Americans and whites with NSCLC to determine whether a difference in *EGFR* mutation frequency exists by race.

## METHODS

Tumors from participants in previous population-based case-control studies of lung cancer from 1985 to 2008 (A.G.S.) who had consented to allow their tissue to be used for research purposes were selected to determine *EGFR* mutation status. These subjects had detailed demographic and information (age, race, smoking status, pack years of smoking) available from previous interviews, and clinical information (histology, stage, and grade) available from the Metropolitan Detroit Cancer Surveillance System, part of the national Surveillance, Epidemiology, and End Results program. Never smokers, light smokers, and individuals with adenocarcinoma were oversampled, and cases were fre-

quency matched by race on sex, smoking status, and histology. Never smokers were defined as individuals smoking fewer than 100 cigarettes in their lifetime.

We used the OncoCarta Panel V1.0 developed by Sequenom, which examined 238 mutations in 19 different oncogenes. This method was chosen because of its sensitivity (as low as 10% mutant allele frequency) and high throughput. For the purposes of this analysis, we focused on 43 known mutations in EGFR included in this panel. Mutation analysis was performed using the mass-spectroscopy based MassArray device developed by Sequenom. DNA was extracted from formalin-fixed paraffin embedded tumors using a standard kit (Qiagene, 51306). Briefly, an initial polymerase chain reaction is performed to amplify a small region, which included the potential point mutation site. Next, a 10-base DNA oligonucleotide primer binds immediately upstream of the mutation site and is extended by one base into the potential mutation site. The primers are subsequently separated on a matrix-assisted laser desorption/ionization time of flight mass spectrometer, which is able to quantitatively discern the specific nucleotide that was extended.

Differences by race and mutation status were assessed for categorical variables using $p$ values from $\chi^2$ tests or Fisher's exact test when expected cell frequency was less than 5. Comparisons were made using $t$ tests for continuous variables. Differences were considered to be statistically significant at alpha $<0.05$. All analyses were performed using SPSS Version 18.

## RESULTS

Tumor samples were obtained from 67 African American and 77 white patients with NSCLC. The demographic characteristics of the patients are listed in Table 1. At the time of diagnosis, African Americans were older compared with whites ($p = 0.03$), but other key parameters did not differ between racial groups. *EGFR* mutations were found in 13.9% of tumors from the total population (Table 2). Fifteen mutations occurred in never smokers with an additional two mutations found in patients with a smoking history of only one pack year. The majority of mutations were found in adenocarcinomas ($n = 18$), with two squamous cell carcinomas also harboring a mutation ($p = 0.10$). The mutation frequency in never smokers was 47% (data not shown). *EGFR* mutations did not differ by race with a frequency of 11.9% in African Americans and 15.6% in whites ($p = 0.53$). Figure 1 illustrates the relationship between smoking status, race, and frequency of different mutation types. Mutations were found in 7 of 11 tumors from African American never smokers (63.6%) and 8 of 21 (38.1%) tumors from white never smokers ($p = 0.27$). All eight mutations in African Americans were exon 19 deletions, whereas mutations in whites included exon 19 deletions (n = 7, 58.3%), the L858R point mutation in exon 21 (n = 3, 25%), and 2 exon 18 mutations, E709A and G719S (16.7%). There was no statistically significant difference between the types of mutations carried by race ($p = 0.17$, data not shown).

**TABLE 1.** Demographic Characteristics of the Study Population, by Race

| | Whites ($n = 77$) *N* (%) | African Americans ($n = 67$) *N* (%) | *p* |
|---|---|---|---|
| Mean age at diagnosis | 55.2 | 60.0 | 0.03 |
| Sex | | | 0.20 |
| Male | 19 (24.7) | 23 (34.3) | |
| Female | 58 (73.3) | 44 (65.6) | |
| Smoking status | | | 0.12 |
| Never | 21 (27.3) | 11 (16.4) | |
| Ever | 56 (72.7) | 56 (83.6) | |
| Histology | | | 0.75 |
| Adenocarcinoma | 57 (74.0) | 48 (71.6) | |
| Other NSCLC | 20 (26.0) | 19 (28.4) | |
| Stage at diagnosis | | | 0.47 |
| I | 30 (39.0) | 20 (29.9) | |
| II | 31 (40.3) | 29 (43.3) | |
| III | 13 (16.9) | 15 (22.4) | |
| Missing | 3 (3.8) | 3 (4.4) | |

NSCLC, non-small cell lung cancer.

**TABLE 2.** Patient Characteristics by EGFR Mutation Status

| | EGFR Mutations ($n = 20$) *N* (%) | Wild Type ($n = 124$) *N* (%) | *p* |
|---|---|---|---|
| Ethnicity | | | 0.53 |
| White | 12 (60.0) | 65 (52.4) | |
| African American | 8 (40.0) | 59 (47.6) | |
| Mean age at diagnosis | 63.8 | 56.4 | 0.02 |
| Sex | | | 0.33 |
| Male | 4 (20.0) | 38 (30.6) | |
| Female | 16 (80.0) | 86 (69.4) | |
| Smoking status | | | <0.001 |
| Never | 15 (75.0) | 17 (13.7) | |
| Ever | 5 (25.0) | 107 (86.3) | |
| Histology | | | 0.10 |
| Adenocarcinoma | 18 (90.0) | 87 (70.2) | |
| Other NSCLC | 2 (10.0) | 37 (29.8) | |
| Stage at diagnosis | | | 0.18 |
| I | 3 (15.0) | 47 (37.9) | |
| II | 10 (50.0) | 50 (40.3) | |
| III | 5 (25.0) | 23 (18.5) | |
| Missing | 2 (10.0) | 4 (3.2) | |

EGFR, epidermal growth factor receptor; NSCLC, non-small cell lung cancer.

## DISCUSSION

Lung cancer remains the leading cause of cancer-related death in the United States with estimates of more than 150,000 deaths in 2009, and an overall 5-year survival of 16%.[10] The majority of these cases are NSCLC. EGFR-TKIs have extended survival in some NSCLC patients. Clinical and biological studies have determined that TKI-responders are primarily those with *EGFR* mutations.[3–6] The recently completed phase III IPASS trial reported that the hazard ratio for

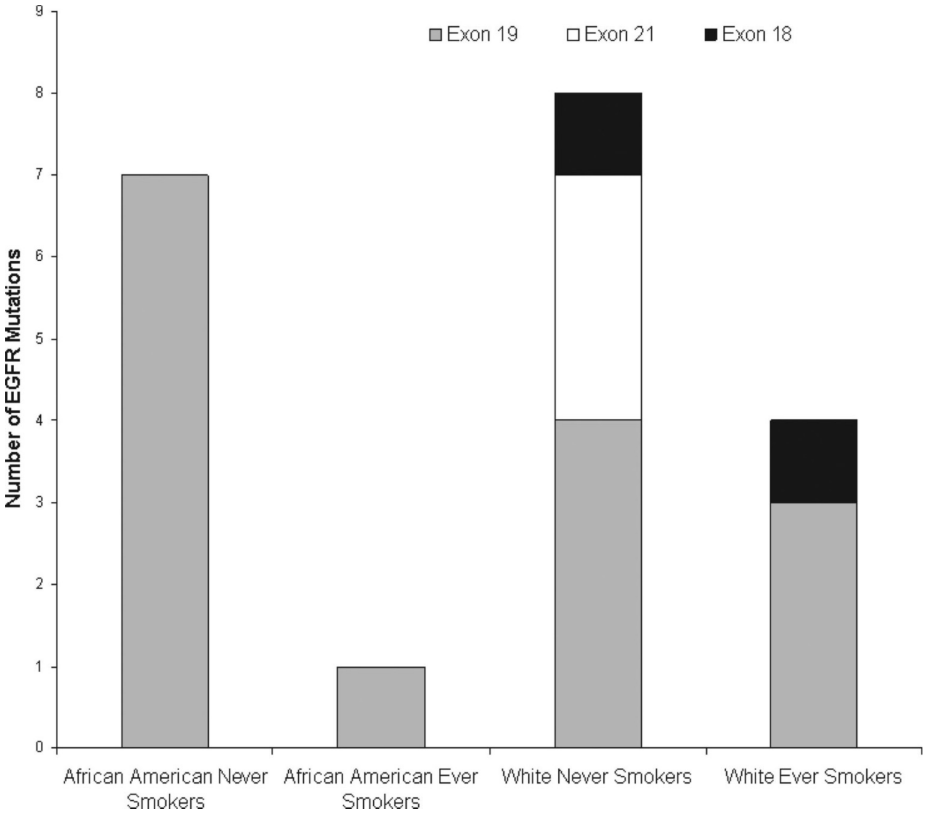


**FIGURE 1.** The number of *EGFR* mutations by race and smoking status.

progression or death was 0.48 in patients with *EGFR* mutations treated with gefitinib compared with those treated with carboplatin-paclitaxel ($p < 0.001$).[1] The 12-month progression-free survival rate was significantly higher with gefitinib in that study of nonsmokers or former light smokers with adenocarcinoma (24.9% in the gefitinib group versus 6.7% for carboplatin-paclitaxel).[1] This study illustrates the clinical relevance of the *EGFR* mutational status for therapeutic decision making in patients with advanced NSCLC.

Contrary to recent reports, we observed no statistically significant difference in *EGFR* mutation frequency in tumor samples from whites and African Americans, in the largest study of African Americans to date. Studies reporting the frequency of *EGFR* mutations, including the present report, must be interpreted with caution because the composition of the patient population may influence the results. *EGFR* mutations occur predominantly, although not exclusively, in never smokers. Pao et al.[5] found *EGFR* mutations in only four of 481 samples from smokers compared with seven mutations in 15 tumors from never smokers. This mutation rate is similar to our observation of 15 mutations in 32 never smokers. Because ~10% of patients with NSCLC are never smokers, *EGFR* mutation rates will be low unless the study population is enriched with these individuals.

The mutation frequency in whites in our study is similar to other reports in white populations.[7–9] In African Americans, Riely et al.[9] studied 14 tumors (eight were never smokers) and found *EGFR* mutations in six patients, similar to our findings that approximately 2/3 of African American never smokers harbored a mutation in *EGFR*. It is unclear why both Yang et al.[7] and Leidner et al.[8] reported finding only a single *EGFR* mutation after studying 41 and 53 African Americans with NSCLC, respectively, but it may have been because of the inclusion of relatively few never smokers in these investigations, or chance.

*EGFR* mutations of importance in NSCLC seem to be confined to the first 4 exons(18–21) of the ATP-binding region of the tyrosine kinase receptor. In our white subjects, exon 19 mutations accounted for two thirds of *EGFR* mutations, yet all the mutations observed in African Americans were deletions in exon 19. Larger studies are needed to determine whether significant differences exist in mutation type between African Americans and whites or whether this is an anomaly related to the sample size or some unique characteristic of our study cohort.

Determination of mutation type may have clinical relevance because some studies have demonstrated that treatment response varies with mutation type. Riely et al.[9] reported that patients with exon 19 deletions treated with TKIs had a median survival of 34 months compared with only 8 months in those with the exon 21 L858R point mutation. Jackman et al.[11] found that length of survival was more than doubled (38 months versus 17 months) for patients with exon 19 deletions. In addition, a study examining the association between *EGFR* mutations and outcome of erlotinib treatment found a higher probability of response associated with the exon 19 deletion ($p = 0.001$).[12] These findings have not been confirmed in two studies involving Japanese patients, in

which no significant difference was noted in progression-free survival based on *EGFR* mutation type.[13,14] In conclusion, this study suggests that *EGFR* mutations occur as frequently in African Americans as in whites. Thus, clinicians should not use race as a clinical decision parameter for the use of EGFR-TKIs.

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